U. S. DEPARTMENT OF AGRICULTURE,
BUREAU OF ANIMAL INDUSTRY.—BULLETIN NO. 86.
A. D. MELVIN, CHIEF OF BUREAU.

EXPERIMENTS WITH MILK ARTIFICIALLY INFECTED WITH TUBERCLE BACILLI.

BY

E. C. SCHROEDER, M. D. V.,
Superintendent of Experiment Station, Bureau of Animal Industry,

AND

W. E. COTTON,
Expert Assistant at Experiment Station, Bureau of Animal Industry.

WASHINGTON:
GOVERNMENT PRINTING OFFICE.
1906.

ORGANIZATION OF THE BUREAU OF ANIMAL INDUSTRY.

Chief: A. D. Melvin.
Assistant Chief: A. M. Farrington.
Chief Clerk: E. B. Jones.
Dairy Division: Ed. H. Webster, chief; Clarence B. Lane, assistant chief.
Inspection Division: Rice P. Steddom, chief; Morris Wooden, assistant chief.
Quarantine Division: Richard W. Hickman, chief.
Animal Husbandman: George M. Rommel.
Editor: James M. Pickens.
Artist: W. S. D. Haines.
Librarian: Beatrice C. Oberly.

LABORATORIES.

Biochemic Division: Marion Dorset, chief.
Pathological Division: John R. Mohler, chief.
Zoological Division: Brayton H. Ransom, scientific assistant in charge.

EXPERIMENT STATION.

E. C. Schroeder, superintendent; W. E. Cotton, assistant.

MEAT INSPECTION.

Inspectors in charge.

Austin, Minn.—Dr. M. O. Anderson, care George A. Hormel & Co.
Baltimore, Md.—Dr. H. A. Hedrick, 215 St. Paul street.
Bloomington, Ill.—Dr. Frederick Braginton, care Continental Packing Company.
Boston, Mass.—Dr. J. F. Ryder, 141 Milk street.
Brightwood, Mass.—Dr. W. J. Murphy, care Springfield Provision Company.
Buffalo, N. Y.—Dr. B. P. Wende, Live Stock Exchange Building, East Buffalo.
Cedar Rapids, Iowa.—Dr. T. A. Shipley, care T. M. Sinclair & Co.
Chicago, Ill.—Dr. S. E. Bennett, room 316 Exchange Building, Union Stock Yards.
Cincinnati, Ohio.—Dr. A. G. G. Richardson, care Union Stock Yards.
Cleveland, Ohio.—Dr. E. P. Schaffter, care Cleveland Provision Company.
Davenport, Iowa.—Dr. E. L. Bertram, care Henry Kohrs Packing Company.
Denver, Colo.—Dr. W. E. Howe, care Western Packing Company.
Des Moines, Iowa.—Dr. A. B. Morse, care The Agar Packing Company.
Detroit, Mich.—Dr. L. K. Green, care Hammond, Standish & Co.
Eau Claire, Wis.—Dr. G. W. Butler, care Drummond Brothers.
Fort Worth, Tex.—Dr. A. H. Wallace, care Swift & Co.
Hutchinson, Kans.—Dr. J. E. Blackwell, care Hutchinson Packing Company.
Indianapolis, Ind.—Dr. N. C. Sorensen, care Kingan & Co.
Jersey City, N. J.—Dr. Julius Huelsen, care The Jersey City Stock Yard Company.
Kansas City, Kans.—Dr. L. R. Baker, room 338 Live Stock Exchange.
Los Angeles, Cal.—Dr. A. E. Rishel, care Cudahy Packing Company.
Louisville, Ky.—Dr. H. H. George, 507 Johnson street.
Mankato, Minn.—Dr. H. H. Dell, care Macbeth & Gardner.
Marshalltown, Iowa.—Dr. J. O. F. Price, care Brittain & Co.
Mason City, Iowa.—Dr. Robert Jay, care Jacob E. Decker & Sons.
Milwaukee, Wis.—Dr. A. E. Behnke, room 432 Federal Building.
Nashville, Tenn.—Dr. W. B. Lincoln, care Tennessee Packing and Provision Company.
National Stock Yards, Ill.—Dr. J. B. Clancy.
Nebraska City, Nebr.—Dr. W. H. Gibbs, care Morton-Gregson Company.
Newark, N. J.—Dr. Thomas Castor, care Swift & Co., Harrison Station.
New Haven, Conn.—Dr. Albert Long, care Sperry & Barnes.
New York, N. Y.—Dr. H. N. Waller, 109 West Forty-second street.
Ottumwa, Iowa.—Dr. Joshua Miller, care John Morrell & Co.
Philadelphia, Pa.—Dr. C. A. Schaufler, 134 South Second street.
Pittsburg, Pa.—Dr. F. W. Ainsworth, Union Stock Yards.
Portland, Oreg.—Dr. Clarence Loveberry, room 402 Custom-House (new).
Quincy, Ill.—Dr. J. S. Kelly, care Blomer & Michael Co.
St. Louis, Mo.—Dr. J. J. Brougham, care Missouri Stock Yards Company.
San Diego, Cal.—Dr. Robert Darling, care Charles S. Hardy.
San Francisco, Cal.—Dr. George S. Baker, Sixth and Townsend streets.
Seattle, Wash.—Dr. O. B. Hess, care Frye-Bruhn Company.
Sioux City, Iowa.—Dr. G. A. Johnson, Exchange Building.
South Omaha, Nebr.—Dr. Don C. Ayer, Post-Office building.
South St. Joseph, Mo.—Dr. George Ditewig.
South St. Paul, Minn.—Dr. F. D. Ketchum.
Tacoma, Wash.—Dr. E. C. Joss, care Carstens Packing Company.
Topeka, Kans.—Dr. F. L. De Wolf, care Charles Wolff Packing Company.
Waterloo, Iowa.—Dr. T. W. Scott, care The Rath Packing Company.
Wichita, Kans.—Dr. W. N. Neil, care John Cudahy Company.
Worcester, Mass.—Dr. E. P. Dowd, care White, Pevey & Dexter Co.

(Concluded on p. 3 of cover.)

U. S. DEPARTMENT OF AGRICULTURE,

BUREAU OF ANIMAL INDUSTRY.—BULLETIN NO. 80.

A. D. MELVIN, CHIEF OF BUREAU.

EXPERIMENTS WITH MILK ARTIFICIALLY INFECTED WITH TUBERCLE BACILLI.

BY

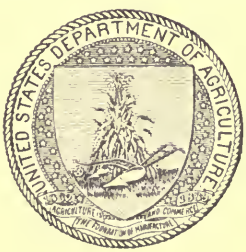
E. C. SCHROEDER, M. D. V.,

Superintendent of Experiment Station, Bureau of Animal Industry,

AND

W. E. COTTON,

Expert Assistant at Experiment Station, Bureau of Animal Industry.



WASHINGTON:

GOVERNMENT PRINTING OFFICE.

1906.

LETTER OF TRANSMITTAL.

U. S. Department of Agriculture,
Bureau of Animal Industry,
Washington, D. C., May 19, 1906.

Sir: I have the honor to transmit herewith, and to recommend for publication as a bulletin in the series of this Bureau, a manuscript entitled "Experiments with Milk Artificially Infected with Tubercle Bacilli," by Dr. E. C. Schroeder and W. E. Cotton, of the Experiment Station of this Bureau. The work consisted of feeding and inoculation experiments with guinea pigs and hogs, and was undertaken to gain more definite information concerning the danger of contracting tuberculosis through the ingestion of milk containing tubercle germs.

This work throws new light upon the tuberculosis problem and is especially important in its bearing on the supposition that the most common way of contracting the disease is through the respiration. The authors conclude that the great frequency of lung tuberculosis need not be wholly ascribed to that form of exposure, but that the lung may and does become infected when the bacilli enter the system in other ways. Special attention is directed to the danger of infection by tuberculous material taken into the body with the food.

Respectfully,

A. D. Melvin,
Chief of Bureau.

Hon. James Wilson,
Secretary of Agriculture.

CONTENTS.

EXPERIMENTS WITH MILK ARTIFICIALLY INFECTED WITH TUBERCLE BACILLI.

AN EXPERIMENT WITH GUINEA PIGS.

Some of the work done recently at the Experiment Station with milk from tuberculous cattle gave results which indicate that the danger of contracting tuberculosis through the ingestion of milk, while it does exist and should be guarded against, is of secondary importance compared with other modes of infection. In order to gain more definite information on the subject, an experiment was made in which a number of guinea pigs were exposed to infection with tuberculosis through milk from normal, healthy cows to which cultures of tubercle bacilli had been added. A detailed account of the experiment follows.

PLAN OF THE EXPERIMENT.

THE INFECTIOUS MATERIAL.

A virulent culture of tubercle bacilli on agar was obtained from the Pathological Division of this Bureau, and a portion of the surface growth was scraped off and suspended in sterile water. The watery suspension contained a sufficient amount of infectious material in a finely divided state to give it a homogeneous, well-marked, cloudy appearance. As much material as would cling to a fine platinum wire loop—the wire about one-fourth mm. and the loop about 2 mm. in diameter—was transferred from the suspension to fresh normal milk at the rate of one loop per 10 c. c. of milk, and this milk was designated "Infection, degree A." From the original watery suspension one loop of material was added to 10 c. c. of sterile water and material from the sterile water to fresh normal milk at the rate of one loop per 10 c. c. of milk, and this milk was designated "Infection, degree B." Again, from the original watery suspension one loop of material was added to 100 c. c. of sterile water and material from the sterile water added to fresh normal milk at the rate of one loop per 10 c. c. of milk, and this milk was designated "Infection, degree C."

During the experiment four separate agar cultures of tubercle bacilli were used, all strongly virulent for guinea pigs, and all made from the same stock culture in the Pathological Division. The cul-

tures were 30 days old at the time they were used, and were received at this station on the following dates, one culture on each day: July 13, July 21, July 27, and August 9. The suspension of tubercle bacilli in water obtained from each culture was used daily to infect the milk, as before described, until a fresh culture was received, when a fresh watery suspension was made and the old one discarded.

METHODS OF INFECTING THE ANIMALS.

Milk of each of the three degrees of infectiousness was used for injecting and feeding, some of the guinea pigs being injected, intra-abdominally, some fed one day, and some fed thirty days.

Before the feeding was begun the guinea pigs were deprived of food and drink for a period of twenty-four hours in order to induce them to take a large quantity of the infected milk. The guinea pigs fed one day received nothing but infected milk after the period of starvation until they had each consumed 50 c. c., and the guinea pigs fed thirty days were deprived of all drink but infected milk during the whole period and consumed an average of 60 c. c. each per day. The average daily amount of milk taken by the 30-day guinea pigs is large, because the little animals soon learn to like milk very much and then drink it eagerly the moment it is placed before them.

The amount of infected milk used for injecting was 5 c. c. for each guinea pig.

Cover-glass preparations from milk of "Infection, degrees B and C," were examined under the microscope, and the number of bacilli in the same was found to be too small for their detection, even when the cover glasses were prepared from the sediment in the bottoms of tubes. The latter were of 15 c. c. capacity and had been rotated in a centrifugal machine for half an hour at the rate of 2,000 revolutions per minute.

RESULTS OF FEEDINGS AND INJECTIONS.

The results obtained from the feedings and injections are shown in the accompanying tables.

TABLE I.—*Results of injections and feedings of milk "Infection, degree A" (1 loop of cloudy suspension in water of tubercle bacilli per each 10 c. c. of milk).*

No. of guinea pig.	Injected or fed.	Date of feeding or injection.	Total amount received.	Date of death.	Autopsy.
			c. c.		
6053	Injected	July 13, 1905	5	Died Aug. 7, 1905..	Generalized tuberculosis.
6054	...do....	.....do.....	5	Died Aug. 5, 1905..	Do.
6079	...do....	July 21, 1905	5	Died Aug. 10, 1905..	Do.
6080	...do....	.....do.....	5	Died Aug. 15, 1905..	Do.
6101	...do....	July 29, 1905	5	Died Sept. 5, 1905...	Do.
6102	...do....	.....do.....	5	Died Sept. 8, 1905...	Do.
6119	...do....	Aug. 9, 1905	5	Died Aug. 31, 1905..	Do.
6120	...do....	.....do.....	5	Died Sept. 8, 1905...	Do.
6025	Fed.....	July 13, 1905	50	Killed Oct. 21, 1905..	No lesions of disease.
6026	...do....	.....do.....	50	.....do.....	Do.
6027	...do....	.....do.....	50	.....do.....	Generalized tuberculosis.
6028	...do....	.....do.....	50	.....do.....	No lesions of disease.
6029	...do....	.....do.....	50	.....do.....	Do.
6030[a]	...do....	.....do.....	50	.....do.....	Do.
6133	...do....	Aug. 11, 1905	50	Killed Oct. 25, 1905..	Do.
6134	...do....	.....do.....	50	.....do.....	Extensive tuberculosis.
6135	...do....	.....do.....	50	.....do.....	No lesions of disease.
6136	...do....	.....do.....	50	.....do.....	Do.
6137	...do....	.....do.....	50	.....do.....	Extensive tuberculosis.
6138	...do....	.....do.....	50	.....do.....	Do.
6013	...do....	July 13 to Aug. 12, 1905	1,800	Died Sept. 13, 1905..	Generalized tuberculosis.
6014	...do....	.....do.....	1,800	Killed Oct. 19, 1905..	Extensive tuberculosis.
6015[a]	...do....	.....do.....	1,800	.....do.....	Do.
6016	...do....	.....do.....	1,800	.....do.....	Do.
6017	...do....	.....do.....	1,800	.....do.....	Do.
6018	...do....	.....do.....	1,800	.....do.....	Do.

[a] Guinea pigs Nos. 6015 and 6030 each produced two young during the experiment, which on autopsy were found to be free from disease.

TABLE II.—*Results of injections and feedings of milk "Infection, degree B" (1 loop of cloudy suspension in water of tubercle bacilli to 10 c. c. of sterile water and 1 loop of the latter per each 10 c. c. of milk).*

No. of guinea pig.	Injected or fed.	Date of injection or feeding.	Total amount received.	Date of death.	Autopsy.
			c. c.		
6051	Injected.	July 13, 1905	5	Died Sept. 27, 1905..	Generalized tuberculosis.
6052	...do....	.....do.....	5	Killed Oct. 21, 1.05..	Do.
6077	...do....	July 21, 1905	5	Died Sept. 7, 1905...	Do.
6078	...do....	.....do.....	5	Killed Oct. 21, 1905..	Do.
6099	...do....	July 29, 1905	5	.....do.....	Do.
6100	...do....	.....do.....	5	.....do.....	Do.
6117	...do....	Aug. 9, 1905	5	.....do.....	Do.
6118	...do....	.....do.....	5	.....do.....	Do.
6031	Fed.....	July 13, 1905	50	Killed Oct. 20, 1905..	No lesions of disease.
6032	...do....	.....do.....	50	Died July 15, 1905...	Pneumonia.
6033	...do....	.....do.....	50	Killed Oct. 20, 1905.	No lesions of disease.
6034	...do....	.....do.....	50	.....do.....	Do.
6035	...do....	.....do.....	50	Died Aug. 14, 1905..	Inflammation of bowels.
6036	...do....	.....do.....	50	Killed Oct. 20, 1905..	No lesions of disease.
6127	...do....	Aug. 11, 1905	50	Killed Oct. 25, 1905..	Do.
6128	...do....	.....do.....	50	.....do.....	Do.
6129	...do....	.....do.....	50	.....do.....	Do.
6130	...do....	.....do.....	50	.....do.....	Do.
6131[a]	...do....	.....do.....	50	.....do.....	Do.
6132	...do....	.....do.....	50	.....do.....	Do.
6043	...do....	July 13 to Aug. 12, 1905	1,800	Killed Oct. 20, 1905.	Do.
6044	...do....	.....do.....	1,800	.....do.....	Do.
6045	...do....	.....do.....	1,800	.....do.....	Do.
6046	...do....	.....do.....	1,800	.....do.....	Do.
6047	...do....	.....do.....	1,800	.....do.....	Do.
6048	...do....	.....do.....	1,800	.....do.....	Do.

[a] One young produced in pen, which on autopsy showed no lesions of disease.

TABLE III.—*Results of injections and feedings of milk "Infection, degree C" (1 loop of cloudy suspension in water of tubercle bacilli to 100 c. c. of sterile water and 1 loop of the latter per each 10 c. c. of milk.)*

No. of guinea pig.	Injected or fed.	Date of injection or feeding.	Total amount received.	Date of death.	Autopsy.
			c. c.		
6049	Injected.	July 13, 1905	5	Killed Oct. 21, 1905..	Extensive tuberculosis.
6050	...do....	.....do.....	5	Died Oct. 5, 1905....	Generalized tuberculosis.
6075	...do....	July 21, 1905	5	Died Oct. 2, 1905....	Do.
6076	...do....	.....do.....	5	Killed Oct. 21, 1905..	Extensive tuberculosis.
6097	...do....	July 29, 1905	5	.....do.....	Do.
6098	...do....	.....do.....	5	.....do.....	Do.
6115	...do....	Aug. 9, 1905	5	.....do.....	Slight tuberculosis.[a]
6116	...do....	.....do.....	5	.....do.....	Extensive tuberculosis.
6019	Fed.....	July 13, 1905	50	.....do.....	No lesions of disease.
6020	...do....	.....do.....	50	.....do.....	Do.
6021	...do....	.....do.....	50	.....do.....	Do.
6022	...do....	.....do.....	50	.....do.....	Do.
6023	...do....	.....do.....	50	.....do.....	Do.
6024	...do....	.....do.....	50	.....do.....	Do.
6121	...do....	Aug. 11, 1905	50	Killed Oct. 25, 1905..	Do.
6122	...do....	.....do.....	50	.....do.....	Do.
6123	...do....	.....do.....	50	.....do.....	Do.
6124	...do....	.....do.....	50	.....do.....	Do.
6125	...do....	.....do.....	50	.....do.....	Do.
6126	...do....	.....do.....	50	.....do.....	Do.
6037	...do....	July 13 to Aug. 12, 1905	1,800	Killed Oct. 19, 1905..	Do.
6038	...do....	.....do.....	1,800	.....do.....	Do.
6039	...do....	.....do.....	1,800	.....do.....	Do.
6040[b]	...do....	.....do.....	1,800	.....do.....	Do.
6041	...do....	.....do.....	1,800	.....do.....	Do.
6042	...do....	.....do.....	1,800	.....do.....	Do.

[a] Tuberculosis limited to one gland near stomach.
[b] Two young produced in pen, which on autopsy showed no lesions of disease.

An examination of the tables shows that all the guinea pigs injected with the infected milk contracted tuberculosis, and hence that every 5 c. c. of the milk at all degrees of infectiousness actually contained live, virulent tubercle bacilli.

Of the guinea pigs that were fed the milk which contained the largest amount of infectious material, 100 per cent of those fed thirty days and 33⅓ per cent of those fed one day became affected with tuberculosis, showing conclusively that the particular tubercle culture used to infect the milk possessed a sufficient degree of pathogenic virulence to cause tuberculosis in guinea pigs through ingestion.

THE SUSCEPTIBILITY OF GUINEA PIGS TO TUBERCULOSIS.

Milk of degrees of infectiousness B and C failed to produce tuberculosis in a single guinea pig fed with it; the 12 that were fed thirty days and consumed each a total of 1,800 c. c. escaped, as well as those fed only a single day with 50 c. c. each. The amount of infectious material in the milk was of course very much less than in milk A, but it was also probably very much greater than the amount present in naturally infected milk from tuberculous cows whose udders are not affected. In our experience with milk from tuberculous cows we have never found a cow with a healthy udder, no matter how extensively she was otherwise affected with tuberculosis, whose milk, on intraabdominal injection of guinea pigs, produced tuberculosis with the regularity

and certainty of our artificially infected milk. From this we may conclude that the milk feeding practiced in this experiment constitutes quite a severe test of the danger encountered by guinea pigs through the ingestion of milk from tuberculous cows whose udders are unaffected. Hence it follows that the ingestion by guinea pigs of milk from tuberculous cows is a very unsatisfactory test for the presence of tubercle bacilli. This is surprising to us, because of the commonly existing belief that guinea pigs are extremely susceptible to tuberculosis, irrespective of the manner in which they are exposed to the infection.

Unfortunately no conclusion can be drawn from the results obtained with the fed guinea pigs of the danger encountered by man through the use of milk from tuberculous cattle, and the failure of the guinea pigs to contract tuberculosis after swallowing innumerable tubercle bacilli suspended in milk should not be construed as an encouragement to use the milk of a cow which is known or even suspected to be affected with tuberculosis. Intestinal tuberculosis of guinea pigs is a very rare occurrence, even when they are affected with otherwise generalized tuberculous disease and every other organ is practically saturated with tuberculous material. It may be that some peculiarity exists about the stomach and intestine of a guinea pig that allows the bacilli of tuberculosis to pass through and out of the body more freely than through the intestines of other species of animals, including man. This view is strengthened by results recently obtained in some hog-feeding experiments at this station in cooperation with the Pathological Division, which will be reported in due time. At this time it is sufficient to say that the hogs in question, on exposure similar to and no more severe than that received by the guinea pigs, readily became affected with tuberculosis.

DISTRIBUTION OF LESIONS IN THE GUINEA PIGS.

The location of the lesions found on post-mortem examination of the 10 guinea pigs that contracted tuberculosis through the ingestion of infected milk warrants a few general remarks. A complete autopsy record of each guinea pig is not required. The following table shows the distribution of the lesions in each animal. Taking the number 9 to represent the total amount of disease found in each guinea pig, the numerals in the table represent approximately the amount of disease found in the several organs. The highest numeral in connection with any guinea pig not only indicates that the organ under which it is placed showed the most extensive tuberculous changes, but that it was, so far as this could be determined, the first organ to become affected. Aside from the amount of disease in any one organ as compared with any or all the other organs in the same animal, the numerals

are not intended to convey any impression as to the actual magnitude of the lesions.

TABLE IV.—*Distribution of the lesions in 10 guinea pigs affected with tuberculosis through the ingestion of infected milk.*

No. of guinea pig.	Throat lymph glands.	Thoracic glands.	Lung.	Portal glands.	Liver.	Spleen.	Mesenteric glands.	Subcutaneous lymph glands.	Intestine.
6013.....	1	1	1	1	1	1	1	1	1
6014.....	5	1	1	1	0	½	½	0	0
6015.....	2	2	½	1	1	½	1	1	0
6016.....	4	2	2	¼	¼	¼	¼	0	0
6017.....	½	1½	1½	1½	1½	2	0	½	0
6018.....	3	1	2	2	½	½	0	0	0
6027.....	0	5	1	1	1	1	0	0	0
6134.....	1	5	1	½	½	1	0	0	0
6037.....	4	3	½	½	½	½	0	0	0
6138.....	0	3	2	1	1	1	1	0	0

The remarkable fact about the distribution of the lesions in the guinea pigs is that, notwithstanding the entrance of the infection with the food, careful search failed to discover lesions of the intestine except in one case, in which practically every organ was affected. The mesenteric lymph glands were affected in only 5 of the 10 guinea pigs, and in no case were the lesions of these glands as important as those found in other portions of the body of the same animal. The subcutaneous glands, under which name the glands in the inguinal and axillary regions are included, were affected in 3 animals; that is, more frequently than the intestine, as the result of ingested infection, although these glands are in no direct manner associated with the organs of digestion.

The liver was affected in 9 animals, but in no case seriously. The portal glands, thoracic glands, lung, and spleen were affected in every guinea pig; and the throat glands, although entirely free from determinable disease in 2 cases, were affected in 8 cases, and 4 of them showed the most marked lesions, being probably the first organs attacked. The animals which did not show the most extensive lesions in the throat glands invariably showed the greatest amount of disease in the thoracic glands.

These facts are interesting mainly because the infection of the animals was strictly through the food that was eaten by them. While the infected food had to pass through the mouth and throat to reach the stomach, its contact with these parts was comparatively of short duration, and its contact with the stomach and bowels of comparatively long duration; therefore more extensive disease of the latter and the associated lymph glands was to be expected.

A SUBCUTANEOUS-INOCULATION EXPERIMENT WITH HOGS.

With reference to the distribution of lesions, the subcutaneous inoculation of 12 hogs with tubercle bacilli is quite interesting. For these inoculations tubercle cultures from four different sources were

used. All the inoculations were made in the central portion of the abdominal region, immediately under the skin, just in front of the navel. The hogs used were part of a number that had been previously used in hog-disease investigations, and were for that reason unsalable and unserviceable for most other purposes, but there was nothing in their condition to unfit them for this experiment.

The tubercle cultures used were obtained from the Pathological Division of this Bureau and were as follows: (1) A culture isolated from a human lung, third generation; (2) a culture isolated from the lung of a boy, a supposedly bovine-human culture, fifteenth generation; (3) a culture isolated from the lung of a hog that had been infected with bovine tuberculosis, twenty-second generation, and (4) a culture isolated from a deer, third generation. For convenience the cultures will be referred to as Nos. 1, 2, 3, and 4.

The post-mortem examinations of the hogs were made with the most scrupulous and searching care, and it is doubtful if any lesions, unless they were extremely small and well concealed, escaped detection. The autopsy records give all the lesions found. The presence of tubercle bacilli in every lesion about the nature of which any doubt existed was microscopically demonstrated.

The records of the hogs follow:

HOGS INOCULATED WITH CULTURE NO. 1.

Hogs Nos. 1751, 1790, and 1805 were each inoculated February 13, 1906, with a small amount of the growth from an agar culture of tubercle bacillus No. 1, and were killed thirty-seven days later and examined post-mortem.

Autopsy of hog No. 1751: Weight at time of death, 59 pounds. At the seat of inoculation is an abscess of about 5 mm. diameter, which contains a dry, firm, cheesy material. The subcutaneous tissues surrounding the abscess in a band less than 3 mm. wide are sprinkled with minute necrotic foci. Lung contains a few small pearl-like tubercles, 2 mm. and less in diameter, uniformly distributed.

Autopsy of hog No. 1790: Weight at time of death, from 55 to 60 pounds. At the seat of the inoculation is an abscess about 1 cm. in diameter, the wall of which is a heavy, dense neoplastic tissue, inclosing a mass of dry, firm, cheesy material. Lung contains about a score of minute pearl-like tubercles, the largest of which is not more than 2 mm. in diameter. Liver contains one small tubercle not more than 1 mm. in diameter.

Autopsy of hog No. 1805: Weight at time of death, 62 pounds. At the seat of the inoculation is an abscess in all respects similar to that found in the same region in hog No. 1751. Lung evenly sprinkled with innumerable pearl-like tubercles, which vary in size from mere points to 2 mm. in diameter. Liver contains a few tuberculous foci, 2 mm. and less in diameter.

HOGS INOCULATED WITH CULTURE NO. 2.

Hogs Nos. 1754, 1755, and 1798 were each inoculated February 13, 1906, with a small amount of growth from an agar culture of tubercle bacillus No. 2, and were killed thirty-seven days later and examined post-mortem.

Autopsy of hog No. 1754. Weight at time of death, from 55 to 60 pounds. At the seat of the inoculation is an abscess about 1 cm. in diameter, which contains a dry, firm, cheesy material. The subcutaneous tissues surrounding the abscess in a band not more than 5 mm. wide are sprinkled with minute necrotic foci. Lung evenly sprinkled with innumerable pearl-like tubercles, the largest of which are 2 mm. in diameter. Liver contains a few tubercles 1 mm. and less in diameter. Spleen contains a very small number of tubercles from 1 to 2 mm. in diameter.

Autopsy of hog No. 1755. Weight at time of death, 45 pounds. Lesions at the seat of the inoculation similar to that found in hog No. 1754. Inguinal lymph glands slightly enlarged and contain several necrotic tuberculous areas from 2 to 3 mm. in diameter. Prescapular lymph glands slightly enlarged and contain several necrotic tuberculous areas from 2 to 3 mm. in diameter, the number of affected areas slightly greater than in the inguinal glands. Lung evenly sprinkled with innumerable minute pearl-like tubercles, the largest of which are 2 mm. in diameter. Bronchial lymph glands enlarged and contain a small number of tuberculous areas. Liver evenly sprinkled with innumerable very minute tubercles. Portal lymph glands contain a small number of minute tubercles. Spleen contains several tubercles from 1 to 3 mm. in diameter.

Autopsy of hog No. 1798. Weight at time of death, 62 pounds. Lesion at the seat of the inoculation similar to that found in hog No. 1754. Lung uniformly sprinkled with numerous pearl-like tuberculous nodules from 1 to 2 mm. in diameter. Liver contains a few minute tubercles.

HOGS INOCULATED WITH CULTURE NO. 3.

Hogs Nos. 1783, 1803, and 1811 were each inoculated February 13, 1906, with a small amount of growth from an agar culture of tubercle bacillus No. 3, and were killed thirty-eight days later and examined post-mortem.

Autopsy of hog No. 1783. Weight at time of death, 55 pounds. At the seat of the inoculation is an abscess about 1 cm. in diameter, which contains a dry, firm, cheesy material. The subcutaneous tissues surrounding the abscess in a band not more than 5 mm. wide are sprinkled with minute necrotic foci. Lung evenly sprinkled with innumerable minute, pearl-like tubercles 2 mm. and less in diameter. Liver contains a considerable number of very minute tubercles. Portal lymph glands contain a few very minute tuberculous foci.

Autopsy of hog No. 1803. Weight at time of death, 52 pounds. At the seat of the inoculation is a lesion similar to that found in hog No. 1783, but not more than one-half as large. Inguinal lymph glands enlarged and some of them contain a small number of necrotic areas 4 mm. and less in diameter. Lung evenly sprinkled with innumerable tuberculous nodules from 1 to 4 mm. in diameter. Bronchial lymph glands enlarged and thickly sprinkled with minute necrotic foci. Liver evenly sprinkled with numerous tubercles, the largest of which are 3 mm. in diameter. Portal lymph glands enlarged and sprinkled with many necrotic foci from 1 to 2 mm. in diameter. Spleen contains a few tuberculous foci from 1 to 5 mm. in diameter. Lymph glands at the curvature of stomach enlarged and sprinkled with necrotic foci from 1 to 2 mm. in diameter.

Autopsy of hog No. 1811. Weight at time of death, 55 pounds. At the seat of the inoculation is a lesion similar in all respects to that found in hog No. 1783. One of the inguinal lymph glands contains a few minute tuberculous foci. Lung evenly sprinkled with numerous pearl-like tubercles 2 mm. and less in diameter. Bronchial lymph glands greatly enlarged and sprinkled with tuberculous foci, some of which have a diameter of 2 mm. Liver contains many tubercles from 1 to 3 mm. in diameter. Portal lymph glands contain several minute foci of tuberculosis. Spleen contains several tuberculous foci from 1 to 3 mm. in diameter.

HOGS INOCULATED WITH CULTURE NO. 4.

Hogs Nos. 1772, 1801, and 1809 were each inoculated February 13, 1906, with a small amount of growth from an agar culture of tubercle bacillus No. 4, and were killed thirty-eight days later and examined post-mortem.

Autopsy of hog No. 1772. Weight at time of death, 46 pounds. At the seat of the inoculation is an abscess about 5 mm. in diameter, which contains a dry, firm, cheesy material. The subcutaneous tissues surrounding the abscess in a band less than 3 mm. wide are sprinkled with minute necrotic foci. Lung contains many pearl-like tubercles 2 mm. and less in diameter located principally in the apexes of the various lobes. Liver contains a few small tubercles, the largest of which are 2 mm. in diameter.

Autopsy of hog No. 1801. Weight at time of death, 55 to 60 pounds. At the seat of the inoculation is a lesion similar to that found in hog No. 1772, but about twice as large. Lung evenly sprinkled with numerous tuberculous nodules from 1 to 3 mm. in diameter. Liver contains a few very small tubercles.

Autopsy of hog No. 1809. Weight at time of death, 61 pounds. At the seat of the inoculation is a lesion precisely similar to that found in hog No. 1801. Lung evenly sprinkled with pearl-like tubercles from 1 to 2 mm. in diameter. Liver contains a few minute tubercles.

DISTRIBUTION OF LESIONS IN THE HOGS.

In the following table the distribution of the lesions in each hog is shown and an attempt is made to represent the proportionate amount of the disease in the affected organs by numerals. It should be stated (as was the case with the guinea pigs in Table IV) that the number placed under each organ is only intended to show the proportion of the disease in that organ as compared with the other organs of the individual hog and has no bearing on the actual total amount of disease present in the hog.

The distribution of the disease in each hog is estimated on a scale of 10. When the number 9, for example, is placed in the lung column, 0.75 in the liver, and 0.25 in the spleen column, it means that the magnitude of the lesions in the lung is nine times as great as in the liver and spleen combined—that is, 9 parts of a total of 10, and the lesions in the liver and spleen would be, respectively, 0.75 part and 0.25 part of 10. The table of course is only approximately correct, because no effort was made to isolate and weigh and measure the lesions in the various organs of the hogs.

The last column of the table shows approximately the magnitude of the total lesions found in each hog as compared with any other hog. In estimating the numbers given the actual mass of the tuberculous tissue present in the body was taken into consideration quite independent of its location or distribution, excepting that the lesions found immediately at the seat of the inoculation were omitted from the estimate. For example, if the total lesions found in hog No. 1751 are 0.1 and the total lesions found in hog No. 1755 are 10 (their respective values in the table) then the actual relation of the volume

of lesions in the one hog to the other is as 0.1 to 10 or 1 to 100. It may give additional value to the figures to state that the actual volume of the tuberculous tissue found in hog No. 1751 (0.1) is equal to about 5 cu. mm.

TABLE V.—*Distribution of lesions in hogs injected with tubercle cultures.*

No. of hog.	Material inoculated.	Inguinal lymph glands.	Prescapular lymph glands.	Lung.	Bronchial lymph glands.	Liver.	Portal lymph glands.	Spleen.	Lymph glands at curvature of stomach.	Relative amount of disease.
1751	Tubercle bacillus No. 1 (human lung).	.....	.....	10.00	.....	.....	.....	.....	.....	0.10
1790		.....	.....	9.90	.....	0.10	.....	.....	.....	1.00
1805		.....	.....	9.90	.....	.10	.....	.....	.....	5.00
1754	Tubercle bacillus No. 2 (bovine-human).	.....	.....	9.00	.....	.75	.....	0.25	.....	5.00
1755		0.25	0.25	5.00	0.25	3.75	0.25	.25	.....	10.00
1798		.....	.....	9.90	.....	.10	.....	.....	.....	2.00
1783	Tubercle bacillus No. 3 (bovine-hog).	.....	.....	9.00	.....	.75	.25	.....	.....	5.00
1803		.75	.....	6.00	.25	2.25	.25	.25	0.25	10.00
1811		.25	.....	5.00	.50	3.50	.25	.50	.....	8.00
1772	Tubercle bacillus No. 4 (deer).	.....	.....	9.50	.....	.50	.....	.....	.....	1.00
1801		.....	.....	9.75	.....	.25	.....	.....	.....	1.00
1809		.....	.....	9.50	.....	.50	.....	.....	.....	1.00

If we now examine the autopsy records of the hogs as they are presented in the foregoing table, we see that tubercle cultures Nos. 2 and 3 are somewhat more virulent for hogs than cultures Nos. 1 and 4. This difference in virulence is not associated with a tendency for the lesions produced by any culture to localize themselves differently from the lesions produced by any other culture.

SUSCEPTIBILITY OF THE LUNG TO INFECTION OTHER THAN BY RESPIRATION.

It will be seen by Table V that the lung was in all the hogs the principal, and very probably the first, organ to become affected, and this certainly could not have been due to the point at which the infectious material was inoculated. The point of inoculation was specially selected to prevent as far as possible the more immediate exposure of some one organ and was located much closer to the liver and spleen than to the lung. The lung disease found was strictly within the lung, some of it under the pulmonary pleura, but not any of it on the pleura of either the lung or the chest wall, and among the twelve affected lungs only three showed an affection of the lymph glands associated with the lung or contained within the thoracic cavity. This condition is not regarded as showing a special affinity of tubercle bacilli for the lung. It is probably due to the fact that the tubercle germs taken up from the subcutaneous tissues successfully passed through the lymph channels and various lymph glands and reached the venous circulation and were then filtered out by the lung. Whether this view is true or not we have in this experiment a demon-

stration of the fact that the location of the tuberculous disease in the body is not necessarily a guide as to the point at which the infectious material entered.

TESTS SHOWING HOW THE LUNG FILTERS THE BLOOD.

The very effective manner in which the lung filters out solid particles from the blood was shown at the experiment station by the injection into the veins of several animals of a carefully prepared suspension in water of pure lampblack. While the specific gravity of lampblack is much greater than that of water, in the ordinary form in which the dry substance can be purchased it is so charged with air or other gases that it is impossible to mix it with water. To overcome this difficulty we heated the lampblack in a crucible to a red heat and put it into water while hot, and found that fully 75 per cent of the mass sank to the bottom at once.

Two rabbits were placed under the influence of ether and 1 c. c. of a suspension of about 10 per cent, by weight, of lampblack in water was injected into their ear veins. The result was almost instantaneous death. Two more rabbits without being placed under the influence of ether were injected in the same manner with the same result: Ether was used in the first instance because it was thought the little animals would suffer long and severely as a result of the injection. It was omitted in the second instance because the operation, followed by almost instantaneous death, caused no more suffering than is endured by exposure to ether during the time that precedes loss of consciousness. The autopsies of the four rabbits showed identical lesions—a uniform distribution of the injected lampblack throughout the lungs—and no trace of the intensely black, very fine, and practically impalpable powder in any other portion of the body.

A horse was also given an intravenous injection of lampblack suspension. In this case the injection was made into the jugular vein and the amount of material in proportion to the weight of the animal was much smaller than that injected into the rabbits. The dose was intentionally made small (50 c. c. of a 1 per cent suspension) so as not to greatly affect the horse, which was an old, vicious animal, not serviceable for other experimental purposes and not safe for ordinary work. The actual amount of lampblack introduced into the circulation of each rabbit was 100 milligrams ($1\frac{1}{3}$ grains) and into the horse 500 milligrams ($7\frac{1}{3}$ grains). As the horse weighed about five hundred times as much as each rabbit the dose it received per weight of animal was one one-hundredth of that received by the rabbits.

The only ante-mortem effect of the injection into the horse was an increased rapidity of the respiration, which lasted several hours and then subsided. About a week after the injection the horse was killed and examined post-mortem. The autopsy revealed the presence of an

even distribution of the lampblack in the lung and no trace of the substance in any other portion of the body. The particles of lampblack in some portions of the lung were very fine and could not be seen without magnification.

Similar evidence of the property of the lung to filter the blood has been observed in the various intravenous injections of tubercle bacilli into cattle that have been made from time to time at the Experiment Station. Dead tubercle germs or tubercle germs of a virulence too low to cause a progressive tuberculous disease when injected into the veins of cattle invariably lodge in the lung and nowhere else where it has been possible to discover them, and cause a miliary tuberculosis from which, if the germs are either dead or very attenuated, recovery is slowly made.

SPREAD OF INFECTION TO OTHER ORGANS.

Next to the lung the organ most frequently affected is the liver, which was affected in 11 cases out of 12, and in 6 of these cases it is the only organ in addition to the lung in which the affection had made its appearance. Following its appearance in the liver we find the disease spreading with equal rapidity to the spleen, portal lymph glands, and bronchial lymph glands, and from then on a tendency to rapid generalization is shown.

If the disease in the liver followed that in the lung, which is almost certain in the twelve injections with which we are dealing, it is more likely to have received the infectious material from the lung than from the seat of the inoculation. Infectious material from the lung we believed entered the circulation through the pulmonary veins and was carried in the blood to the heart and thrown into the arterial circulation and filtered out when the blood reached the liver, the peculiar circulation of which may specially enable it to act as a very efficient natural filtering system.

REMARKS ON THE HOG INOCULATIONS.

There are some facts that must be kept in mind in estimating the practical significance of the course followed by the tuberculous disease in this group of hogs. First, the affection was produced by the introduction of a number of bacilli vastly greater than is likely to occur with a natural exposure; hence the number of individual original foci of disease that developed simultaneously was vastly greater than from a natural infection. Second, there is a condition known as immunity to tuberculosis that can be produced by either the intravenous or the subcutaneous injection of tubercle bacilli of a pathogenic virulence somewhat less than is required to cause an active, progressive tuberculosis.

The first fact probably caused the presence of many more germs in the blood of the hogs at all times after the inoculation than occurs in

the case of a naturally acquired tuberculosis, and a deposition of these germs first of all in the lung, then in the liver, then in the spleen, and later in other structures, from which they were taken up by the lymph glands.

In a naturally acquired tuberculosis, that begins with possibly one or two or at most a few foci of disease in one organ, the other organs of the body have time to acquire some immunity similar to that referred to as the second fact. Such immunity would play a very small part in protecting the organs of the inoculated pigs, because of the intense character of the exposure and the rapid spread of the disease. We have in the hogs a strictly acute tuberculosis, and wish to distinguish this from the chronic form in which the affection is usually encountered in nature among men and other animals. This probably explains the absence of lymph-gland disease in 8 of the 12 hogs. In the acute disease we have the distribution of infectious material going on through the circulation; in the chronic disease we have the increase progressing about the peripheral portions of each focus and, because of their incessant exposure, an early affection of the lymph glands that first receive the drain from the tissues in which the lesions are located; and when in the chronic disease an occasional bacillus is carried by the blood to the liver or spleen the acquired resistance—the immunity that may be caused in some organs by the presence of tuberculosis in other organs—prevents the development of fresh disease in most instances. This view receives some support from the fact that animals, for example, with extensively tuberculous lungs swallow an amount of infectious material many times greater than healthy animals can obtain anywhere on natural exposure without necessarily causing tuberculous disease of the throat glands or abdominal organs. The ingestion of infectious material is evidently much more dangerous to healthy animals than to the still unaffected organs of animals already affected with tuberculosis, and if the records of the guinea pigs in the first portion of this article have any value they show that ingested tuberculous material is more apt to cause disease of the throat and lungs than of any structure contained in the abdominal cavity.

NEW SIGNIFICANCE OF LUNG INFECTION.

The supposition that tuberculosis of the lung is commonly due to respired infection is shown to be unnecessary to account for the presence of the disease more frequently in the lung than in other portions of the body. The lung, it seems, is most commonly affected simply because the lymph collected from all portions of the body, after it reaches the blood circulation, must pass through the lung before it reaches any other organ, and the lung evidently will not admit of the passage of solid particles, other than the elastic blood corpuscles,

through its filtering net of capillaries. The result is that it makes no difference at what point the tuberculous material enters the body there is nothing to protect the lung from infection but the lymph glands, and that the lymph glands frequently allow tuberculous material to pass through them or by them without becoming affected is conclusively shown by the series of hog inoculations.

The inoculations of the hogs were superficial—in the middle of the abdominal region—and the lung was affected in 100 per cent of cases, and in only 25 per cent of cases was any disease present in the lymph glands in any portion of the thoracic cavity. In 58⅓ per cent of cases the disease was confined entirely to the lung and liver, without disease of any lymph gland in the body.

Of course there may be some difference between the anatomical structure of the lymph glands of hogs and other animals of a kind that will allow the easier passage of bacteria through the former, and likewise similar differences in the structure of the lymph glands located in different portions of the body of the same animal. But that does not greatly militate against the conclusion that the lung is more directly exposed to tuberculous affection, respired affection left out of consideration, than any other organ.

Respired infectious material comes to rest in the lung on the mucous surface of the bronchial tubes and is then still located on what may be regarded as one of the exterior surfaces of the body. In this location, because of the irritation produced by the material with which it gains entrance—dust, etc.—it has an excellent chance to become enveloped with mucous secretions and to be coughed up and either swallowed or expectorated. This consideration, taken in connection with the facts we have presented to account for the more frequent presence of the disease in the lung than elsewhere, seems to show that tuberculosis due to bacilli that enter the lung with the breathed air is an uncommon affection. If this conclusion is true, its practical significance lies in the caution to be doubly sure that our food and drink is free from tubercle germs, and especially that it is wise to avoid the use of the milk produced by tuberculous cows or in stables containing tuberculous cattle.

RESISTANCE OF TUBERCLE GERMS.

The tubercle bacillus is a peculiar organism in its conduct in the animal body, independent of its real pathogenic significance. When cultures of other bacteria are injected into the body of an animal insusceptible to the affection of which the germs in question may be the specific cause, these germs or bacteria disappear entirely from the body in a very short time. The same is not true of the bacillus of tuberculosis; on the contrary, it shows an enormous resistance to the destroying and annihilating processes that are brought to bear

against it by the body. Even dead or sterile cultures of tubercle bacilli injected into cattle or sheep remain lodged in the tissue for months and years in a manner in which their presence can be microscopically demonstrated.

SUMMARY.

To sum up, we believe the experiments presented have shown the following facts:

1. That the high susceptibility of guinea pigs to tuberculosis holds good only when the infectious material is introduced into the body in a way in which it can not escape through the natural excretory organs; that is, when it is injected under the skin, into the abdominal cavity, into the veins, into the thorax, etc.

To strengthen this conclusion, we may add that we recently exposed 52 guinea pigs and 6 hogs to tuberculosis through milk feeding, the milk given the guinea pigs and the hogs being indentical in its infectious character. The result was that 5 of the 6 hogs contracted tuberculosis and the 52 guinea pigs remained unaffected and in perfect health. A more detailed report of this experiment will be published at another time.

2. That the localization of tuberculous disease in the lung of an animal gives us no information as to the point at which the infectious material entered.

3. That the lung is more especially and directly exposed to tuberculous affection than any other organ, because of the character of its circulation and because the entire lymph stream that is poured into the circulation must pass through the lung before it reaches the capillary structures or smaller and finer vessels of any other organ.

4. That it is not necessary to account for the great frequency with which tuberculosis localizes itself in the lung by supposing that the most common form of exposure to tuberculosis is through the respiration.

5. That the experiments, taken as a whole, direct special attention to the danger sustained through exposure to tuberculous material that enters the body with the food. This fact can not be too strongly emphasized.

O

Inspectors in charge of districts.

Dr. R. A. Ramsay, room 320 Quincy Building, Denver, Colo., in general charge of eradication of scabies of sheep and cattle in the West.
Albuquerque, N. Mex.—Dr. Louis Metsker, room 22 N. T. Armijo Building.
Denver, Colo.—Dr. Lowell Clarke, room 320 Quincy Building.
Fargo, N. Dak.—Dr. R. H. Treacy.
Kansas City, Kans.—Albert Dean, room 328 Live Stock Exchange.
Salt Lake City, Utah.—George S. Hickox, room 21 Eagle Block.

INSPECTION OF LIVE STOCK FOR EXPORT.

Inspectors in charge.

Baltimore, Md.—Dr. H. A. Hedrick, 215 St. Paul street.
New York, N. Y.—Dr. W. H. Rose, 18 Broadway.
Norfolk, Va.—Dr. G. C. Faville, P. O. box 796.
Philadelphia, Pa.—Dr. C. A. Schaufler, 134 South Second street.
Portland, Me.—Dr. F. W. Huntington, U. S. customs office, Grand Trunk R. R. wharf.

INSPECTION AND QUARANTINE OF IMPORTED ANIMALS.

Quarantine stations.

Athenia, N. J. (for the port of New York).—Dr. George W. Pope, superintendent.
Halethorp, Md. (for the port of Baltimore).—William H. Wade, superintendent.
Littleton, Mass. (for the port of Boston).—Dr. J. F. Ryder, inspector in charge, 141 Milk street, Boston, Mass.

Inspectors on the Canadian border.

Calais, Me.—Dr. H. T. Potter.
Carthage, N. Y.—Dr. W. S. Corlis.
Detroit, Mich.—Dr. L. K. Green, care Hammond, Standish & Co.
Fort Fairfield, Me.—Dr. F. M. Perry.
Malone, N. Y.—Dr. H. D. Mayne.
Newport, Vt.—Dr. G. W. Ward.
Ogdensburg, N. Y.—Dr. Charles Cowie.
Orono, Me.—Dr. F. L. Russell.
Port Huron, Mich.—Dr. David Cumming, 912 Lapeer avenue.
St. Albans, Vt.—Dr. C. L. Morin.
Sault Ste. Marie, Mich.—Dr. J. F. Deadman.

Inspectors on the Mexican border.

El Paso, Tex.—Dr. Thomas A. Bray.
San Antonio, Tex.—Dr. Joseph W. Parker.
San Diego, Cal.—Dr. Robert Darling, care Charles S. Hardy.

VETERINARY INSPECTORS STATIONED ABROAD.

Dr. W. H. Wray, 34 Streatham Hill, London, S. W., England, in charge for Great Britain and Ireland.
Dr. T. A. Geddes, care U. S. consulate, London, England.
Dr. V. A. Nörgaard, Honolulu, Hawaii.

DAIRY INSPECTORS.

M. W. Lang, room 423 Marine Building, Chicago, Ill.
Robert McAdam, room 423 Marine Building, Chicago, Ill.
E. A. McDonald, Seattle, Wash.
Levi Wells, Laceyville, Pa.
G. M. Whitaker, Washington, D. C.